HDL-C and HDL-C/apoA-I predict long-term progression of glycemia in established type 2 diabetes.

Short: HDL-C, Apo-Al and Glycemia

Boris Waldman, BSc*1; Alicia J. Jenkins, MD*1; Timothy M.E. Davis, MRCP2; Marja-Riitta Taskinen, MD3; Russell Scott, PhD FRACP4; Rachel L. O’Connell, PhD1; Val J. Gebski, MStat1; Martin K.C. Ng, PhD#5; Anthony C. Keech, FRACP#1 on behalf of the FIELD Study investigators.

*Alicia Jenkins and Boris Waldman are joint first authors of this study.
# Martin Ng and Anthony Keech are joint senior authors of this study.

1. NHMRC Clinical Trials Centre, University of Sydney, Sydney; Australia.
2. School of Medicine, University of Western Australia, Fremantle; Australia.
3. Department of Medicine, University of Helsinki, Helsinki, Finland.
4. Lipid & Diabetes Research Group, Christchurch Hospital, Christchurch, New Zealand.
5. Department of Cardiology, Royal Prince Alfred Hospital, Sydney, Australia.

Corresponding authors:
Boris Waldman
Research Fellow
Boris.waldman@ctc.usyd.edu.au

or
Anthony Keech
Deputy Director
NHMRC Clinical Trials Centre
tony@ctc.usyd.edu.au

Locked Bag 77
Camperdown NSW 1450
Sydney, Australia
Abstract

Objective: Low HDL-C and HDL particle size may directly promote hyperglycemia. We evaluated associations of HDL-C, apoA-I, and HDL-C/apoA-I with insulin secretion, insulin resistance, HbA1c and long-term glycemic deterioration, reflected by initiation of pharmacologic glucose control.

Research Design and Methods: The 5-year FIELD study followed 9795 type 2 diabetes subjects. We calculated baseline associations of fasting HDL-C, apoA-I and HDL-C/apoA-I with HbA1c and, in those not taking exogenous insulin (n=8271), with estimated beta-cell function (HOMA-B) and insulin resistance (HOMA-IR). Among the 2608 subjects prescribed lifestyle only, Cox proportional hazards analysis evaluated associations of HDL-C, apoA-I and HDL-C/apoA-I with subsequent initiation of oral hypoglycemic agents (OHAs) or insulin.

Results: Adjusted for age and sex, baseline HDL-C, apoA-I and HDL-C/apoA-I were inversely associated with HOMA-IR (r = -0.233, -0.134, -0.230, all p<0.001, n=8271) but not related to HbA1c (all p>0.05, n=9795). ApoA-I was also inversely associated with HOMA-B (r=-0.063, p=0.002, n=8271) adjusted for age, sex and HOMA-IR. Prospectively, lower baseline HDL-C and HDL-C/apoA-I levels predicted greater uptake (per 1-SD lower: HR=1.13 [CI: 1.07–1.19], p<0.001; HR=1.16 [CI: 1.10 – 1.23]; p<0.001) and earlier uptake (median 12.9 months and 24.0 months respectively for quartile 1 versus quartile 4; both p<0.005) of OHAs and insulin, with no difference in HbA1c thresholds for initiation (p=0.87, p=0.81). Controlling for HOMA-IR and triglycerides lessened both associations, but HDL-C/apoA-I remained significant.
Conclusions: HDL-C, apoA-I and HDL-C/apoA-I were associated with concurrent insulin resistance but not HbA1c. However, lower HDL-C and HDL-C/apoA-I predicted greater and earlier need for pharmacologic glucose control.
Introduction

Type 2 diabetes is a progressive disease, characterized by insulin resistance and ongoing loss of endogenous insulin secretion with increased requirement for pharmacologic glucose control over time (1, 2). Low HDL cholesterol (HDL-C) is a common finding in type 2 diabetes patients and is best known as a predictor of cardiovascular risk (3-5). Furthermore, changes to HDL-C levels, HDL particles and their major apolipoprotein, apoA-I, are reported to be present years before the development of type 2 diabetes (6-12). This raises the question of whether HDL biology contributes directly to the development of type 2 diabetes and the continuing progression of the disease.

Associations between HDL related measures and incident type 2 diabetes may reflect co-morbid conditions such as hypertriglyceridemia, abdominal obesity and insulin resistance, which also portend incident disease. However, recent pre-clinical evidence suggests that the action of HDL particles and apoA-I can independently promote insulin secretion and glucose uptake in patients with type 2 diabetes (13, 14). Human islet-cell culture and animal studies have reported that exogenous HDL improves insulin secretion through increased reverse cholesterol transport and attenuation of LDL and inflammation-induced apoptosis of pancreatic beta-cells (13, 15). HDL and apoA-I may also promote glucose uptake by skeletal muscle through activating the AMPK pathway (16). In humans with type 2 diabetes, measures of endogenous HDL function have been inversely associated with concurrent estimated beta-cell function (17). Also, infusion of exogenous reconstituted HDL over 4 hours increased both insulin secretion and skeletal muscle glucose uptake in a small trial with type 2 diabetes patients (18). Cross-sectional studies have reported an inverse association between HbA1c and HDL-C in type 2 diabetes (19, 20). Further, a number of studies have linked HDL-C levels to the development of incident type 2 diabetes (6-8, 10). However, to
date, no study has examined the relationship between these biomarkers and the progression of established diabetes.

The Fenofibrate Intervention and Event Lowering in Diabetes (FIELD) trial was a randomized controlled trial of fenofibrate therapy in 9795 adult type 2 diabetes patients, with measures of HDL-C and apoA-I at baseline. We investigated the whether HDL-C, apoA-I and HDL particle size, as estimated by the HDL-C/apoA-I ratio (21), were inversely associated both cross-sectional measures of glycemia and prospectively with requirements for escalation of glucose control therapies, which would be consistent with HDL having anti-diabetic effects. We also considered associations of the HDL-related measures with two key determinants of glycemia, pancreatic beta-cell secretion and insulin resistance.
Research Design and Methods

Subjects
The FIELD study design has been published previously (22, 23). All participants had an initial total plasma cholesterol of 3.0–6.5 mmol/L, plus a total cholesterol to HDL-C ratio of 4.0 or higher, or a fasting plasma triglyceride reading of 1.0–5.0 mmol/L. The study excluded those who had an existing indication for lipid modifying therapy, renal impairment (plasma creatinine ≥ 130 micromol/L), chronic liver disease (alanine aminotransferase (ALT) ≥ 2 times the upper limit of normal), symptomatic gallbladder disease, or those who had experienced a cardiovascular event within the 3 months before recruitment. Patients were followed up every 4 months in the first year and then every 6 months for a median of 5 years.

Measurements and outcomes
Fasting serum HDL-C, serum apoA-I, serum insulin and HbA1c were measured at baseline in two core laboratories aligned to the Canadian Reference Laboratory. We then calculated the HDL-C/apoA-I ratio. In subjects not taking exogenous insulin (n=8271), Homeostasis Model Assessment (v2.2.2, 12 December 2007; www.dtu.ox.ac.uk) was used to derive estimates of pancreatic β-cell secretion (HOMA-B) and insulin resistance (HOMA-IR) from fasting plasma glucose and serum insulin (24). As recommended by the model’s authors, HOMA calculations were further limited to patients with glucose readings between 3 and 25 mmol and insulin levels between 20 and 400 pmol (25). Medications were recorded at each visit. Baseline adiposity was assessed using BMI, waist circumference and waist to hip ratio. Renal function was assessed using both estimated GFR and the presence of albuminuria. Evidence of fatty liver change was inferred from alanine aminotransferase (ALT) readings. Self-reported smoking status, regular alcohol consumption and physical exercise were recorded at baseline. At all centers, changes in medication use were recorded at
baseline and at each subsequent visit by a qualified clinical trials nurse using a common drug
dictionary which was later converted to ATC code. For patients commencing the trial on
lifestyle measures alone, we regarded initiation of oral hypoglycemic agents or insulin
therapy by the patients’ usual general practitioner or endocrinologist, once confirmed, as
evidence of escalation of diabetes therapy.

Statistical analysis
We first considered age and sex-adjusted partial correlations between baseline variables,
which were logarithmically transformed as appropriate. Partial correlations with HOMA-B
were additionally adjusted for HOMA-IR. Linear regression analysis was used in order to
further adjust for possible confounders, which were selected based on known or suspected
association with the variables of interest. All baseline analyses were repeated in the lifestyle
only sub-group (n = 2608) to avoid possible confounding effects of OHAs or insulin therapy.
In the same sub-group, we used Cox proportional hazards models to consider whether lower
HDL-C, apoA-I or HDL-C/apoA-I was associated with more rapid uptake of OHAs and
insulin therapy during follow up. Interaction terms for treatment allocation were evaluated.
Models were adjusted for HOMA-IR, BMI and HbA1c, triglycerides, waist circumference,
hypertension, LDL-C, ALT, eGFR, the presence of albuminuria, current smoking, regular
alcohol consumption, physical exercise and menopause status in women. In separate time-
dependent Cox regression analyses we evaluated the effect of uptake of statins and
angiotensin converting enzyme inhibitors (ACEi) / angiotensin receptor blockers (ARBs)
during follow-up. Where a HDL-related measure was found to be a significant predictor of
progression to pharmacologic glucose control, we aimed to exclude possible interactions
between the measure and glycemic management. We classed participants into sex-stratified
quartiles according to each relevant measure. Using a one-way ANOVA, we assessed
whether the highest quartile of the measure was associated with a greater rise in HbA1c than
the lowest quartile. In those who initiated pharmacologic glucose control, we compared the last recorded HbA1c before therapeutic progression between the highest and lowest quartiles of a measure, using a Kruskal-Wallis test. P-values are presented unadjusted for multiple comparisons. All analyses used SPSS 20.0 (IBM Corp., Armonk, NY) or SAS 9.1 software (SAS Institute Inc., Cary, NC).

Results

The 9795 FIELD participants had a mean age of 62±7 years with a median diabetes duration of 5 years (IQR 2-10 years), median HbA1c of 6.8% (IQR 6.1-7.8%) [51 mmol/mol (IQR 43–62 mmol/mol)] and median HDL-C of 1.10 (0.27) mmol/L. We recorded Caucasian ethnicity for 93% of participants. The 2608 participants in the lifestyle only subgroup tended to have a shorter duration of diabetes (2.0 v 5.0 yrs), a lower HbA1c (6.0 v 6.8%) [(42 v 51 mmol/mol)] and a higher HOMA-B score (64.2 v 50.2) than other participants in the FIELD cohort but were otherwise comparable. HOMA-B and HOMA-IR values were calculated for 8271 participants in the general cohort and 2560 participants in the lifestyle only subgroup. Baseline characteristics for the entire cohort as well as for subgroups on hypoglycemic pharmacotherapy and on lifestyle-measures only are presented in Table 1.

Baseline age and sex adjusted partial correlations are shown in Table 2. There were no significant partial correlations of HDL-C, apoA-I or HDL-C/apoA-I with HbA1c in the whole cohort (n=9795) or in the lifestyle only subset (n=2608). This persisted after additional adjustment, by linear regression, for possible confounders, which were duration of diabetes, adiposity, triglycerides, HOMA-IR, renal function and ALT. HOMA-IR was inversely correlated to HDL-C, apoA-I and HDL-C/apoA-I (r = -0.223, -0.134, -0.230 respectively in the whole cohort (N = 8271), all p<0.001), which persisted following further adjustment for possible confounders. Adjusted for age, sex and HOMA-IR, there was a small inverse partial
correlation between HOMA-B and apoA-I in both the whole cohort and the lifestyle only cohort \((r=-0.041, \ p<0.001; \ r=-0.063; \ p=0.002\) respectively) which persisted following adjustment for BMI, waist-to-hip ratio triglyceride levels, inferred renal function, ALT levels and diabetes duration.

Over a median follow-up period of 5 years, 1520 (58.3%) of the 2608 participants using lifestyle only measures at baseline commenced an OHA or insulin therapy. Cox proportional hazards analysis, adjusted for age and sex, found that lower HDL-C and HDL-C/apoA-I, but not apoA-I, predicted a greater risk of initiation of OHAs or insulin therapy (HDL-C: \(HR = 1.13\ \ [CI: \ 1.07 – 1.19], \ p<0.001; \) apoA-I: \(HR = 1.04\ \ [CI: \ 0.99 – 1.10], \ p = 0.102; \) HDL-C/apoA-I: \(HR = 1.16\ \ [CI: \ 1.10 – 1.23]; \ p<0.001\) per 1-SD lower). As fenofibrate is known to affect HDL metabolism (26), we checked if there was any interaction between the HDL-related variables and treatment allocation. HDL-C and HDL-C/apoA-I were significant predictors of initiation of pharmacologic glucose control in both the fenofibrate and placebo arms of the study separately (HDL-C: \(HR=1.13, \ p=0.003\) (placebo arm) v \(HR=1.12, \ p=0.003\) (fenofibrate arm); HDL-C/apoA-I: \(HR=1.13, \ p=0.002\) (placebo arm) v \(HR=1.21, \ p<0.001\) (fenofibrate arm)) and the interaction terms with treatment allocation were insignificant for both HLD-C (\(p=0.95\)) and HDL-C/apoA-I (\(p=0.19\)). Those in the lowest baseline sex-stratified HDL-C quartile (median 0.83mmol/L) progressed to pharmacologic glucose control a median of 13 months earlier than those in the highest quartile (median 1.40mmol/L) (figure 1). Those in the lowest baseline sex-stratified HDL-C/apoA-I quartile (median 0.28) progressed to pharmacologic glucose control a median of 24 months earlier than those in the highest baseline quartile (median 0.37). Annualized rises in HbA1c did not differ significantly according to baseline HDL-C quartile (0.079 %/year [0.86mmol/mol/yr] Q1 v. 0.073 %/year [0.80mmol/mol/yr] Q4, \(p=0.58\)) or baseline HDL-C/apoA-I quartile (0.076 %/year [0.83mmol/mol/yr] Q1 v. 0.057 %/year [0.62mmol/mol/yr] Q4, \(p=0.10\)). In those who
progressed to pharmacologic glucose control, the median HbA1c values prior to initiation of pharmacologic glucose control did not differ significantly either for baseline HDL-C quartile (7.0% [53mmol/mol] Q1 v. 7.1% [54mmol/l] Q4, p=0.75) or HDL-C/apoA-I quartile (7.0% [53mmol/mol] Q1 v. 7.1% [54mmol/l] Q4, p=0.71). We then adjusted cumulatively for HOMA-IR, BMI and HbA1c, which we had previously reported as determinants of progression to pharmacologic glucose control in part of the FIELD cohort (23) (figure 2). After further adjusting for triglycerides, only HDL-C/apoA-I remained a significant predictor of progression to pharmacologic glucose control over 5 years. Additional adjustment for waist to hip ratio, LDL-C, ALT, eGFR and albuminuria, current smoking, regular alcohol consumption, physical exercise and menopause status in women did not significantly affect either predictor. Neither statin nor ACEi/ARB initiation during follow-up appreciably altered the prospective results (not shown). Similarly, inclusion of data on patients whose HOMA parameters are not recommended to be calculated did not materially change either the cross-sectional or prospective results.
Conclusions

In our large study of predominantly Caucasian subjects with established type 2 diabetes we report the novel observation that lower levels of HDL-C and a lower HDL-C/apoA-I ratio predict substantially earlier initiation of pharmacologic glucose control among those initially treated with lifestyle measures alone. This is in spite of no statistically significant baseline cross-sectional associations between HDL-related measures and HbA1c levels. The inverse association of progression to pharmacologic glucose control with HDL-C/apoA-I, which estimates HDL size, persists after adjustment for multiple metabolic and lifestyle factors. The results are thus consistent with the emerging concept that HDL biology may play a direct role in the development and progression of type 2 diabetes.

Our prospective findings need to be considered in light of our cross-sectional analysis which did not find hypothesized associations of HDL-C, apoA-I or HDL-C/apoA-I with HbA1c or HOMA-B. There was no inverse correlation between HbA1c levels and HDL-related measures, despite low HDL-C being more common in type 2 diabetes patients than in the general population (3). Two smaller studies of type 2 diabetes patients in Italy and Saudi Arabia have previously reported modest but statistically significant inverse associations between HDL-C and HbA1c \( r = -0.183, p<0.05, r = -0.074, p<0.005 \) respectively) (19, 20). In both studies, participants appeared to have higher average HbA1c and HDL-C levels and longer diabetes duration than subjects in our study. Although our cohort is typical of an early type 2 diabetes population it was selected for the purposes of a clinical trial. Lipid values formed part of the selection criteria and thus may have limited variability in HDL-C at baseline. Among all subjects who were screened for the trial \( n=13900; \) HDL-C range 0.29-1.47 v 0.45-2.96 mmol/L in the randomized cohort) there was a small inverse correlation between HDL-C and HbA1c \( r = -0.030, p<0.001, \) age- and sex-adjusted. Furthermore, residual insulin secretion and intensity of glycemic therapy are primary determinants of
concurrent glycemia. Thus important determinants of long-term glycemia may not be reflected in cross-sectional analyses (1). Although insulin resistance is a major determinant of the development and progression of type 2 diabetes (1), HOMA-IR predicted around 1% of the variation in HbA1c at baseline in our cohort ($R^2=0.009$, $p<0.001$).

Our cross-sectional study also found no significant associations of HDL-C, apoA-I or HDL-C/apoA-I with HOMA-B after adjustment for age, sex and HOMA-IR. HOMA-B is positively correlated with HOMA-IR ($r=0.514$, $p<0.001$), since higher insulin resistance drives greater compensatory insulin secretion in early type 2 diabetes (1, 27). Furthermore HOMA-IR is inversely associated with HDL-related measures in the present study. This necessitates adjusting associations of HOMA-B for variation in HOMA-IR (17). A very small inverse association between apoA-I and HOMA-B ($R^2=0.001$; $p=0.001$) emerged after adjustment for HOMA-IR and all other available confounders. Although we cannot explain this association, it appears to be too small to be of clinical relevance. Notably, a cross-sectional study of 22 type 2 diabetes patients found no significant associations between HOMA-B and HDL-C or apoA-I, despite finding that HDL-related reverse cholesterol transport and antioxidant potential were positively associated with HOMA-B (17).

The progressive nature of type 2 diabetes warrants investigation of factors which may influence the rate of glycemic deterioration. The finding that lower HDL-C and HDL-C/apoA-I values portend earlier initiation of pharmacologic glucose control suggests that HDL metabolism may be contributory. For FIELD trial participants treated with lifestyle alone at baseline, the median time from diabetes diagnosis to need for pharmacotherapy was 6 years, a full year earlier among those in the lowest HDL-C/apoA-I quartile, and a full year later for those in the highest quartile, despite similar glycemic control and management throughout the study. The results accord with multiple studies showing that HDL-related measures predict incident type 2 diabetes (6-10, 28). A single smaller study has previously
reported a relationship between HDL-C and initiation of OHAs in established type 2 diabetes. It considered 705 patients who had an HbA1c\(\leq7\%\) [HbA1c\(\leq53\text{mmol/mol}\)] and were using lifestyle measures alone at baseline (29). In unadjusted analyses, baseline HDL-C levels were lower amongst those who progressed to OHAs or whose HbA1c levels rose above 7% at the end of one-year.

Similarly, we have used the progression from lifestyle measures to pharmacologic glucose lowering as an index of worsening glycemia. During the trial, HbA1c below 7.0% [53mmol/mol] was the accepted target and first OHAs were instituted at a median HbA1c of 7.1% [54mmol/mol] in FIELD subjects (23, 30). Nevertheless, the study protocol did not dictate glycemic management hence the possibility would exist that an association between low baseline HDL-C levels and initiation of first glycemic therapy might reflect more aggressive prescribing related to perceived low HDL-C mediated risk. In order to exclude this possibility, we examined whether baseline HDL-C (or HDL-C/apoA-I) levels influenced either the HbA1c levels at which pharmacotherapy was commenced or total HbA1c rises over 5 years. Neither analysis supported such an alternative explanation. Also, lower baseline HDL-C and HDL-C/apoA-I predicted both earlier metformin and sulphonylurea uptake suggesting that our results were independent of the mode of pharmacotherapy employed (results not shown).

Our longitudinal analysis is adjusted for multiple lifestyle and metabolic factors, which could influence relationships between HDL-related measures and type 2 diabetes progression. Low physical activity, alcohol non-consumption and smoking are lifestyle factors that are associated with lower HDL-C and multiple other metabolic alterations (31, 32). These factors did not materially affect the predictive value of HDL-C or HDL-C/apoA-I in our cohort. Similarly, the Prevention of Renal and Cardiovascular End-Stage Disease (PREVEND) study recently reported that both HDL-C and HDL-C/apoA-I predicted incident type 2 diabetes
independently of current smoking and alcohol consumption (8). In the Diabetes Prevention Program (DPP), on-study HDL-C rise was associated with less progression to type 2 diabetes in the intensive lifestyle, metformin and control arms (28).

In contrast to lifestyle factors, metabolic factors, in FIELD, partially account for the observed relationship between HDL-related measures and pharmacologic glucose control initiation. After controlling for HOMA-IR and triglycerides, the hazard ratios for both HDL-C and HDL-C/apoA-I were attenuated and only HDL-C/apoA-I remained a significant predictor of progression to OHAs or insulin. Elevated insulin resistance and triglycerides are both independently and inversely associated with reduced HDL-C and HDL/apoA-I (33). Both factors are also thought to promote islet cell stress and apoptosis via associations with systemic low grade inflammation and increased free fatty acid levels (27, 34, 35). Higher triglyceride levels and larger VLDL particles have also been reported to predict incident type 2 diabetes (9, 10). However studies diverge as to whether HDL-related or triglyceride-related measures are stronger predictors of incident disease. Importantly, the PREVEND study recently reported that both HDL-C and HDL-C/apoA-I predicted incident type 2 diabetes independent of triglyceride levels or HOMA-IR (8).

Our study thus provides further support for the notion that smaller cholesterol-poor HDL particles, reflected by lower HDL-C levels and HDL-C/apoA-I ratios, may contribute directly to worsening glycemia. Alteration to HDL size and cholesterol content may reflect reduced efficacy of reverse cholesterol transport in type 2 diabetes (36, 37). Since HDL-mediated reverse cholesterol transport has been positively associated with insulin secretion and broader anti-inflammatory effects, this could provide a link between a reduced HDL/apoA-I ratio and more rapid deterioration in glycemic control (14, 18, 38).

Strengths of our study include a large sample size, and the ability to account for multiple possible confounders, including lifestyle factors, metabolic factors and statin and ACEi/ARB
use. The length of follow-up and the high rate of therapy initiation enabled us to derive median times to event, which has not been possible in studies of diabetes incidence. The study has some limitations. Caucasian ethnicity was reported by 93% of subjects, which may limit the generalizability of the results. Lifestyle factors, which can influence HDL metabolism, were self-reported and may be affected by under-reporting. Unless under-reporting was differential with respect to HDL, this would only increase random error rather than introducing bias. Recording of escalation of diabetes therapy could contain inaccuracies, however any possible misclassification would result in dilution of potential associations with HDL-related parameters, rather than the converse. This sub-study was not a stated purpose of the FIELD trial, however all analyses were performed after a hypothesis and pre-specified analysis plan were prepared. One of the FIELD inclusion criteria was a total-cholesterol-to-HDL-C ratio greater than 4. This may have some bearing on the range and spread of HDL-related measures in our cross-sectional analysis but, if anything, would have lead us to underestimate our significant longitudinal associations. HOMA model estimates are not as reliable as those using clamp methods, and could not be carried out on all subjects. However they are a useful proxy in the context of a large cohort (39). HDL function and size were not directly measured in this study and HDL-C and apoA-I may not reflect HDL function well (14, 17, 37). However HDL-C is the most studied HDL-related measure and is predictive of cardiovascular and microvascular complications in type 2 diabetes (4, 5, 40). ApoA-I is widely available, and elucidating its predictive value in type 2 diabetes is important. The HDL-C/apoA-I ratio is easy to derive, reasonably approximates HDL particle size, and has been previously linked to incident type 2 diabetes (8, 21). Nonetheless, large clinical studies evaluating the associations of HDL particle number and function with concurrent and long term glycemia in established type 2 diabetes would be of interest.
In summary, lower levels of baseline HDL-C and HDL-C/apoA-I were not cross-sectionally associated with HbA1c, but predicted earlier initiation of pharmacologic glucose control over 5 years in people with pre-existing type 2 diabetes. In particular, the value of HDL-C/apoA-I, an estimate of HDL particle size, in predicting glycemic progression, was independent of all measured confounders. This provides clinical support for a direct involvement of HDL biology in worsening glycemia in people with established type 2 diabetes.
Author Declarations

B.W. designed the current study, conducted the data analysis, interpreted the results and drafted the manuscript. A.J. designed the current study, interpreted the data and drafted the manuscript. T.D. designed and conducted the FIELD study, contributed to data interpretation and edited the manuscript. M.R.T. designed and conducted the FIELD study, contributed to data interpretation and edited the manuscript. R.S. conducted the FIELD study, contributed to data interpretation and edited the manuscript. R.O’C. maintained the FIELD database and conducted statistical analysis on the current study. V.G. supervised data analyses and contributed to interpretation of the results. M.N. designed the current study, contributed to data interpretation and edited the manuscript. A.K. designed and conducted the FIELD study, designed the current study, contributed to the data analysis and interpretation, and the writing of the manuscript. A.K. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Parts of this manuscript were presented as abstracts at the American Heart Association Scientific Sessions 2013 and at the Cardiac Society of Australia and New Zealand Annual Scientific Meeting 2013.

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study had no role in data collection or data analysis. This secondary study did not have specific funding. None of the authors have conflicts of interest to disclose in relation to this sub-study. David Espinoza, NHMRC Clinical Trials Centre, assisted with the statistical validation. The FIELD Investigators thank the Heart Foundation Australia, Diabetes Australia, Diabetes New Zealand and the Finnish Diabetes Association for endorsing the study. B.W. extends his thanks to the Nematallah and Kamle Habib Family Scholarship Fund and the University of Sydney Medical School for their generous support of his studies.
References


Table 1 – Baseline characteristics for participants.

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Lifestyle Measures Only N = 2608</th>
<th>OHAs and Insulin Therapy N = 7187</th>
<th>Entire Cohort N= 9795</th>
<th>( P^* )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male (%)</td>
<td>1616 (62.0%)</td>
<td>4522 (62.9%)</td>
<td>6138 (62.7%)</td>
<td>0.387</td>
</tr>
<tr>
<td>Age (yrs)</td>
<td>62.2 (6.8)</td>
<td>62.2 (6.9)</td>
<td>62.2 (6.9)</td>
<td>0.622</td>
</tr>
<tr>
<td>Diabetes Duration (yrs)</td>
<td>2.0 (1-5)</td>
<td>6 (3-11)</td>
<td>5.0 (2 – 10)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>29.3 (26.4 – 33.0)</td>
<td>30.0 (27.0-33.8)</td>
<td>29.8 (26.7-33.5)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>102 (13)</td>
<td>104 (13)</td>
<td>104 (13)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Waist to hip ratio</td>
<td>0.92 (0.08)</td>
<td>0.94 (0.08)</td>
<td>0.93 (0.08)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Laboratory Data</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HbA1c (%)</td>
<td>6.0 (5.6 – 6.7)</td>
<td>7.2 (6.4-8.2)</td>
<td>6.8 (6.1-7.8)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>HbA1c (mmol/mol)</td>
<td>42 (38-50)</td>
<td>55 (46-66)</td>
<td>51 (43-62)</td>
<td></td>
</tr>
<tr>
<td>Plasma Glucose (mmol/L)</td>
<td>7.1 (6.2 – 8.2)</td>
<td>8.9 (7.4-10.9)</td>
<td>8.3 (6.9-10.3)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Serum Insulin (U/ml)</td>
<td>12 (8.0 – 17)</td>
<td>12 (8.0-19)</td>
<td>12 (8.0 – 18.5)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>LDL-C (mmol/L)</td>
<td>3.16 (0.67)</td>
<td>3.08 (0.66)</td>
<td>3.10 (0.66)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>HDL-C (mmol/L)</td>
<td>1.11( 0.27)</td>
<td>1.09 (0.26)</td>
<td>1.10 (0.27)</td>
<td>0.009</td>
</tr>
<tr>
<td>apoA-I (g/L)</td>
<td>1.30 (0.21)</td>
<td>1.29 (0.21)</td>
<td>1.29 (0.21)</td>
<td>0.028</td>
</tr>
<tr>
<td>HDL-C/apoA-I (mg per mg)</td>
<td>0.33 (0.04)</td>
<td>0.32 (0.04)</td>
<td>0.32 (0.04)</td>
<td>0.020</td>
</tr>
<tr>
<td>Triglycerides (mmol/L)</td>
<td>1.68 (1.28 – 2.21)</td>
<td>1.74 (1.33-2.32)</td>
<td>1.72 (1.32 – 2.29)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Alanine aminotransferase (ALT) (U/L)</td>
<td>22.5 (17.5 – 30.5)</td>
<td>24.0 (18.0-33.0)</td>
<td>23.5 (17.5 – 32.5)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Estimated Glomerular Filtration Rate (eGFR)</td>
<td>88.2 (18.6)</td>
<td>88.0 (19.4)</td>
<td>88.1 (19.2)</td>
<td>0.703</td>
</tr>
<tr>
<td>Microalbuminuria†</td>
<td>555 (21.3%)</td>
<td>1549 (21.6%)</td>
<td>2104 (21.5%)</td>
<td>0.788</td>
</tr>
<tr>
<td>Macroalbuminuria†</td>
<td>97 (3.7%)</td>
<td>307 (4.3%)</td>
<td>404 (4.1%)</td>
<td>0.228</td>
</tr>
<tr>
<td>HOMA derived values‡</td>
<td>N = 2560</td>
<td>N = 5711</td>
<td>N = 8271</td>
<td></td>
</tr>
</tbody>
</table>
Table 1: Continuous data are expressed as median (interquartile range) or mean (SD), and categorical data are shown as indicated. *Statistical tests compared those commencing on lifestyle only measures to those commencing on OHAs and/or insulin therapy. P values were obtained using $\chi^2$ tests for categorical variables, one-way ANOVA for normally distributed continuous variables, or Kruskal–Wallis tests for non-normally distributed continuous variables. †Microalbuminuria is defined as urine albumin-to-creatinine ratio ≥2.5 mg/mmol and <25mg/mmol for men and ≥3.5 mg/mmol and <35 mg/mmol for women. Macroalbuminuria is defined as urine albumin-to-creatinine ratio >25mg/mmol for men and >35mg/mmol for women. ‡ The HOMA model defines HOMA-B = 100% and HOMA-IR= 1 as normal.
Table 2 – Age and sex adjusted associations of baseline HDL-related and glycemic variables (partial correlations).

<table>
<thead>
<tr>
<th></th>
<th>HbA1c</th>
<th>HOMA-IR</th>
<th>HOMA-B</th>
<th>HOMA-B (adjusted for HOMA-IR)*</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Whole Cohort</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N = 9795</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N = 8271</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HDL-C</td>
<td>-.009</td>
<td>-.223^</td>
<td>-.128^</td>
<td>-.016</td>
</tr>
<tr>
<td>apoA-I</td>
<td>.002</td>
<td>-.134^</td>
<td>-.103^</td>
<td>-.041^</td>
</tr>
<tr>
<td>HDL-C/apoA-I</td>
<td>-.001</td>
<td>-.230^</td>
<td>-.100^</td>
<td>.021</td>
</tr>
<tr>
<td><strong>Lifestyle Only</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N = 2608</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N = 2560</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HDL-C</td>
<td>-.029</td>
<td>-.245^</td>
<td>-.174^</td>
<td>-.037</td>
</tr>
<tr>
<td>apoA-I</td>
<td>-.012</td>
<td>-.169^</td>
<td>-.151^</td>
<td>-.063^</td>
</tr>
<tr>
<td>HDL-C/apoA-I</td>
<td>-.018</td>
<td>-.254^</td>
<td>-.131^</td>
<td>.026</td>
</tr>
</tbody>
</table>

HbA1c, HOMA-IR and HOMA-B were logarithmically transformed prior to correlation and regression analyses as described in the methods section.

^a. correlation is significant at the 0.05 level (2-tailed)
^b. correlation is significant at the 0.01 level (2-tailed)
^c. correlation is significant at the 0.001 level (2-tailed)
^*. partial correlation adjusted for age, sex and HOMA-IR.
Figure Legends

Figure 1

Figure 1: Kaplan-Meier curves showing percent of subjects remaining on lifestyle measures alone over time, according to baseline (A) HDL-C and (B) HDL-C/apoA-I quartiles (sex stratified). Quartiles 2 and 3 are combined for clarity. The median values for quartiles 1 and 4 were 0.83mM and 1.40mM for HDL-C and 0.29 and 0.36 for HDL-C/apoA-I respectively. The median times to progression to pharmacotherapy were 13 months and 24 months earlier for quartile 1 than 4 by HDL-C and HDL-C/ApoA1 respectively.

Figure 2

Figure 2: Hazard ratios and 95% confidence intervals for uptake of pharmacologic glucose therapy – results are displayed as the effect of 1 standard deviation (SD) lower baseline value of (A) HDL-C and (B) HDL-C/apoA-I respectively. Results are cumulatively adjusted for age and sex, then HOMA-IR, BMI, HbA1c, Triglyceride levels, and lastly also for the potential confounders of LDL-C, waist circumference, hypertension, ALT, eGFR, the presence of micro or macroalbuminuria, female menopause status, current smoking, regular alcohol consumption and physical exercise.